

A Microscopical Study of the Structure of Meat Emulsions and Its Relationship to Thermal Stability

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ABSTRACT

Meat emulsions were examined by light microscopy to determine the relationships between structure and emulsion stability. Beef-meat based emulsions were prepared either by varying chopping temperature or by adding soy plastic fats of different hardness. Formation of fat channels was observed in uncooked emulsions prepared either at a chopping temperature of 26°C or with soft plastic fat of 0.25cm⁻¹ hardness index. Such fat channel formations caused discontinuity of protein matrix, leading to fat separation during cooking and weakening of textural strength of products. Changes in fat globule size and distribution as affected by melting properties or hardness of fats were clearly reflected by changes in emulsion stability. Results suggest that emulsion stability is determined largely by the physical properties of protein matrix and fat incorporated. Stable emulsions were obtained with fats of appropriate melting properties and hardness and relatively cohesive protein matrix under given comminution conditions which allowed uniform fat distribution.

INTRODUCTION

THE OCCURRENCE of emulsion destabilization is rare in a routine meat packing operation. Nevertheless, emulsion destabilization still remains one of the concerns in the meat processing industry, especially when new sources of protein and ingredients are incorporated or when inevitable changes in the processing system occur. In this situation, stable emulsions may not be obtained under the conventional processing system unless the underlying physical changes are carefully taken into consideration in formulation and processing.

A comminuted meat emulsion may be considered as a gel-type emulsion in which fat is dispersed uniformly in a continuous protein gel matrix. This gel-type emulsion is different in physicochemical properties from an oil-in-water emulsion in which the interfacial film plays a major role in a fat stabilization and the fat droplets always remain globular in a suspension state. The interfacial film is formed as a result of adsorption of protein on the surface of fat globule which is surrounded by a continuous liquid phase (Mita et al., 1974; Schut, 1978; Smith et al., 1980). The stability of oil-in-water emulsions is therefore believed to be influenced primarily by the viscoelasticity of film (Becher, 1955; Bikerman, 1958; Biswas and Haydon, 1962), and can be predicted by the Stokes' Law (Lissant, 1974).

On the other hand, in a gel-type emulsion the fat droplets are physically confined within the protein matrix and thus the shape of the fat droplets does not necessarily remain globular. They may coalesce with each other and form somewhat larger droplets, but they cannot escape from the matrix to produce a single phase. It is thus assumed that stability depends largely on the rigidity of the gel and the distribution of the fat droplets at the beginning of a gel matrix formation.

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Frequent problems have arisen while applying the oil-in-water emulsion principle to a gel-type comminuted meat emulsion system due to discrepancies in the physicochemical nature between these two systems. Compared to the oil-in-water system in which much work has been done, only limited information is available on the physical aspects of a fat stabilization in gel-type emulsions.

Considering the meat emulsions as a particulate composite system, major factors affecting the emulsion stability appear to be physical properties of protein-fat matrix, the fat-protein interaction and the size and distribution of fat droplets.

The present study was initiated to show how these factors affect the thermal stability of gel-type emulsions through examining structure and its relationship to physical properties.

MATERIALS & METHODS

Preparation of emulsions

Meat emulsions were prepared and cooked under different processing conditions which reflect major stability factors. Following the processing scheme as shown in Fig. 1, prechilled ground lean chuck (0°C, 3.2% fat) was chopped with other ingredients in a five-pound capacity Fleetwood bowl chopper (Model FC-14, Lowenstein, Inc.) equipped with twin blades (6 cm diam). Our preliminary microscopic examination showed that the 15-min initial chopping produced a fine protein gel matrix. The clearance between the blade and the bowl was 0.1 cm. Fat was added such that fat contents in the batters remain 22% on the average.

Fat distribution pattern was altered by the following two manners: (1) varying the temperature of the batter prepared with beef trimming fat, and (2) adding soy plastic fats of different hardness. Soybean oil-based plastic fats were prepared by blending an oil and a fully hydrogenated solid fat (Durkee Foods, SCM Corp., Cleveland, OH) in the ratios which produced 10%, 30%, and 50% solid fat contents, respectively. Hardness was determined using a micro-penetrometer and expressed in terms of a reciprocal of penetration distance which was termed as hardness index (HI) (Lee et al., 1980).

Temperature of the batter was varied to 16, 21, and 26°C, respectively, at a given chopping time (total 30 min) by altering the ingredient and the ambient temperature: for 16°C, 0 (ingredient temperature) - 12°C (ambient temperature); for 21°C, 12-21°C; and for 26°C, 12-26°C. Batters prepared as such were stuffed into casings of 23 mm diam (Precision Nojax, Union Carbide

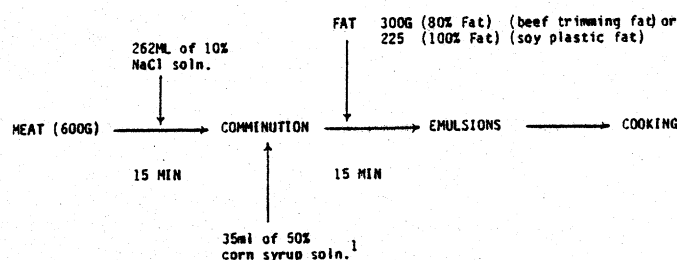


Fig. 1—Preparation of meat emulsions: 1DE 42, Clinton Corn Processing Co., Clinton.

Corp.) and cooked at two different heating rates (Fig. 2) in a smoke-house equipped with temperature and humidity controls.

Measurement of viscosity

The viscosity of the batters was measured at the completion of chopping using a Brookfield viscometer (Model RVT) with a spindle #7 (3mm diam and 5 cm height). Triplicate measurements were made at three rotational speeds, 10, 20, and 50 rpm, respectively. Shear stress (τ , dyne/cm²) at the wall of spindle was computed using an equation, $\tau = F \cdot \%M/R^2 \cdot 2\pi \cdot L$, where F = full torque capacity (7,187 dyne-cm), M = torque reading, R = radius of spindle, and L = height of spindle. Shear rate ($\dot{\gamma}$, sec⁻¹) was calculated using an equation, $\dot{\gamma} = 2\pi R \cdot \text{RPS}/\delta$, where δ = thickness of the portion of batter involved in shearing action. The apparent viscosity (μ , poise) at a given rotational speed was then computed using an equation, $\mu = \tau/\dot{\gamma}$.

Thermal stability

A plug of batter weighing approximately 20g, which was obtained from each preparation, was placed in a 60 ml capacity glass

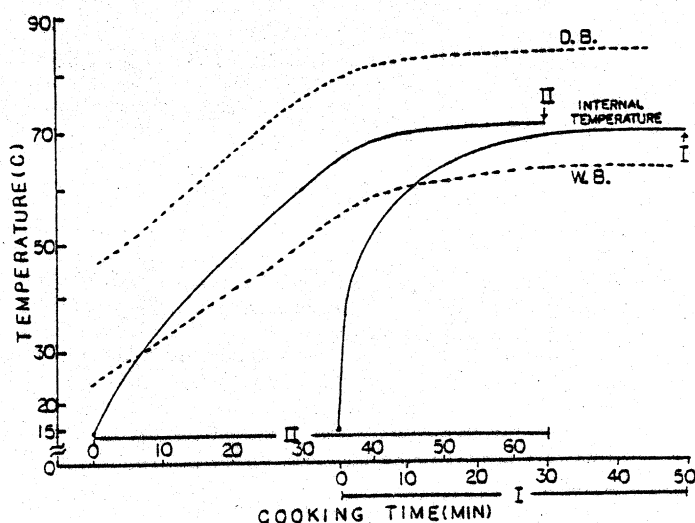


Fig. 2—Temperature history of cooking at two different heating rates: I and II represent fast and slow heating, respectively.

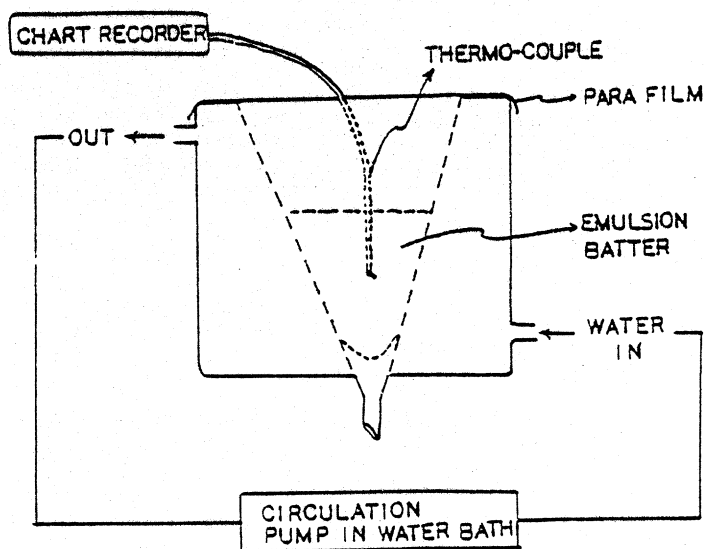


Fig. 3—A set-up for a measurement of commencing temperature: A centrifugal circulating (immersion type) pump was used.

jar and heated in a water bath at 60°C and 70°C, respectively, for 30 min. The amount of fat and water released upon heating was measured by transferring into a 10 ml graduated cylinder in order to determine a retention. The percent retention was used as a thermal stability index. Moisture contents were determined following the AOAC oven drying method (1975). Fat contents was determined following the method of Bligh and Dyer (1959) with some modifications where approximately 10g of sample was homogenized in a 100 ml capacity Waring Blendor for 1 min with 50 ml of a chloroform-methanol mixture (1:1, v/v). The 5 ml of chloroform layer was transferred into a vial and allowed to evaporate. The resulting residue was used as a crude fat residue. The internal temperature of the batter at which a release of fluid commenced was measured by monitoring changes in the internal temperature. As shown in Fig. 3 approximately 20g of batter was placed in a water-jacket funnel and a thermo-couple probe (copper-constantan) was placed in the center of the batter by resting the tip at 1/3 of the specimen height. Temperature of the water passing through the jacket was maintained at either 60° or 70°C by circulating water of equivalent temperature at a rate of 30 ml/sec. The temperature at which the batter yielded the first drop of fluid was used as a commencing temperature.

Microscopical examination of structure

The structure of both cooked and uncooked products was examined by light microscopy. Specimens were frozen in liquid nitrogen and sectioned 16μ thick using a microtome cryostat (IEC Minotome). The sections were then fixed with glutaraldehyde vapor by laying a slide over the 25% glutaraldehyde for 10 min in the hood with a vent on. The sections fixed as such were stained with oil red O for fat globules, subsequently with hematoxylin for protein matrix.

Mechanical properties of protein gel matrix

Mechanical properties of cooked products were evaluated with an Instron Universal Testing Machine (Model 1122) using compression test. Cylindrical specimens of uniform geometry (20 mm ht and 20 mm diam) were tested in five replications at a deformation rate of 50 mm/min and at a chart speed of 100 mm/min. From a single compression (70%) force was measured at failure, while from three cyclic compressions (50%) unrecoverable deformation was measured. A percent plastic deformation was computed using an equation, $(D_3 - D_1 / \text{specimen height}) \times 100$, where D_1 and D_3 are distance between compression head and specimen before the first compression and after the third compression, respectively.

Statistical analysis of the data

The data were analyzed for the statistical significance of differences between treatments by the standard Student's t-test employing the Statistical Analysis System (SAS, 1979).

RESULTS & DISCUSSION

THE PHOTOMICROGRAPHS of uncooked emulsions prepared at three different chopping temperatures are shown in Fig. 4. When the batter was chopped to 16°C, the fat was dispersed uniformly throughout a protein matrix without showing a fat coalescence (Fig. 4-A). As chopping temperature rose gradually, the fat started to soften and coalesce at 21°C (Fig. 4-B) and continued to coalesce, leading to a formation of fat channels as the temperature of the batter reached 26°C (Fig. 4-C). During cooking, the fat was separated from the matrix to the extent of fat coalescence, developing interstitial openings which caused a discontinuity of matrix as seen in the batter chopped to 21°C (Fig. 5-B) and 26°C (Fig. 5-C). Such structural changes during cooking did not occur to the batter which was chopped to 16°C (Fig. 5-A). This indicates that a stability of the protein-fat matrix is determined by the fat dispersion pattern established during comminution. It was also observed that once the localization of fat was completed at the end of chopping, there were no further changes in the distribution patterns during either storage or cooking. Emulsions remained stable through cooking as long as fat was dispersed in a discrete form regardless of size and shape. Chopping to

26°C caused a complete disruption of the protein-fat matrix. This temperature appears to closely lie near the range of the temperatures at which the phase transition of beef trimming fat occurs (28–30°C) (Townsend et al., 1968). Similar fat distribution patterns were observed in emulsions which were prepared at constant temperature (16°C) with soy plastic fats containing 30% (equivalent to 0.87 cm⁻¹ HI) (Fig. 6-a) and 10% solid fat contents (equivalent to 0.25 cm⁻¹ HI) (Fig. 6-b). The results indicate that the meat emulsion may be destabilized not only by increasing chopping temperature beyond the softening point of fat, but also by incorporating soft fat (solid fat content 10%).

When a hard fat (soy plastic fat of 3.0 cm⁻¹ HI or rendered beef kidney fat of 2.1 cm⁻¹ HI) was added to the batter at 26°C, the emulsion remained stable although the fat was not distributed as uniformly as in the emulsion prepared with a medium hard fat (Fig. 7). Considering a significant change in the viscoelastic properties of interfacial film that would occur to the batter at 26°C, the stability of gel-type emulsions may not reflect the changes in the viscoelastic properties of interfacial film due to a temperature rise. Conversely, in an oil-in-water emulsion, the viscoelastic properties of interfacial film plays an important role in a fat stabilization. It is therefore suggested that instead of

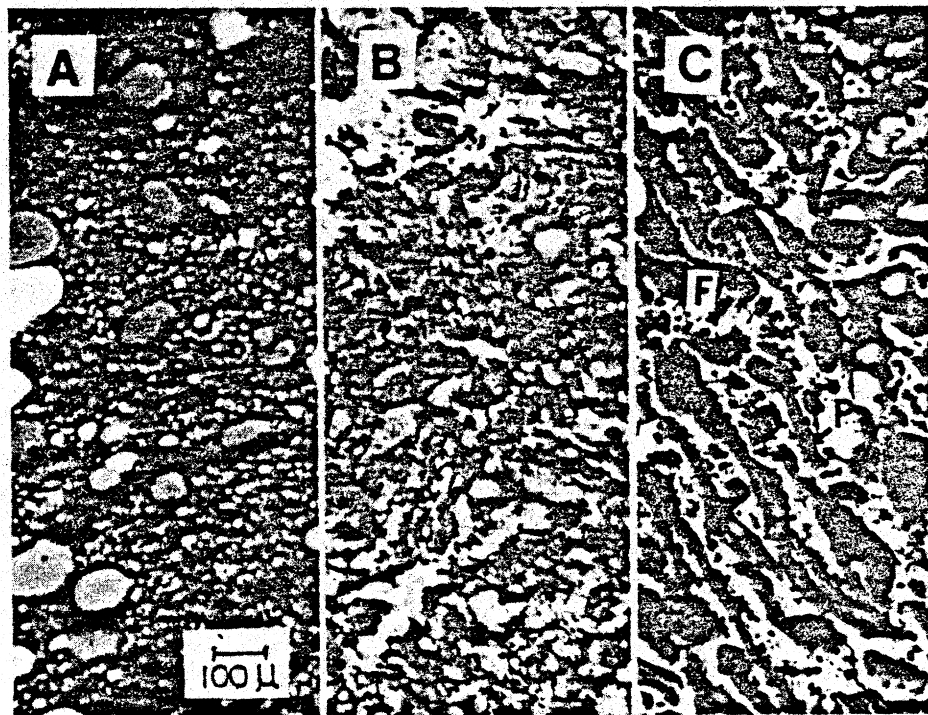


Fig. 4—Photomicrographs of uncooked emulsions at different chopping temperatures: (A) 16°C, (B) 21°C, and (C) 26°C, where F and P represent fat droplets and disintegrated protein matrix, respectively.

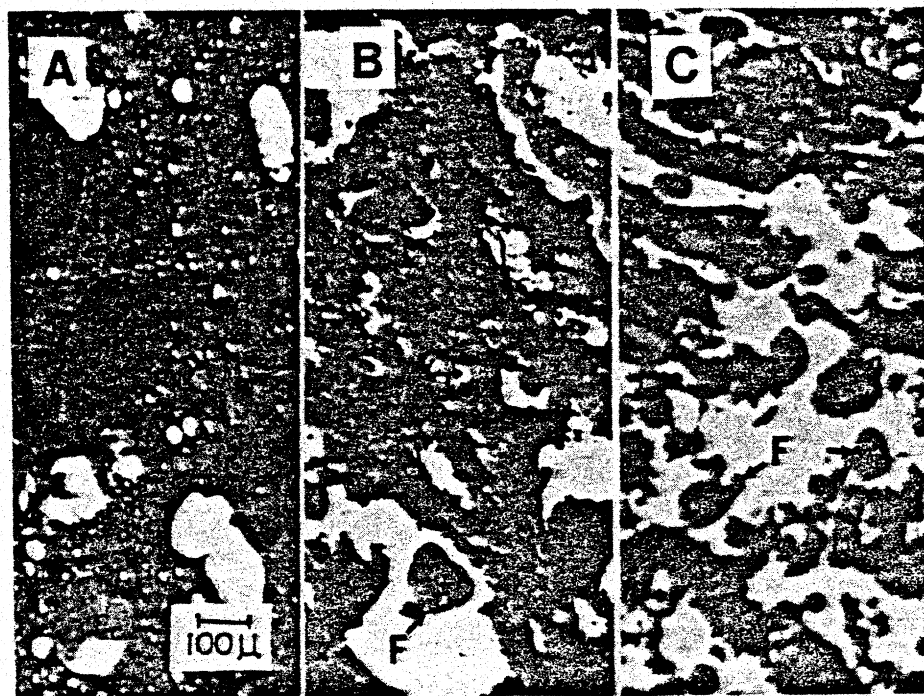


Fig. 5—Photomicrographs of cooked emulsions prepared at different chopping temperatures: (A) 16°C, (B) 21°C, and (C) 26°C, where F represents fat droplets.

Fig. 6—Photomicrographs of emulsions prepared with soy plastic fats of different hardness: Uncooked—*a* ($0.87\text{ cm}^{-1}\text{ H1}$) and *b* ($0.25\text{ cm}^{-1}\text{ H1}$). Cooked—*A* ($0.87\text{ cm}^{-1}\text{ H1}$) and *B* ($0.25\text{ cm}^{-1}\text{ H1}$), where *F*, *P*, and *V* represent fat droplets, protein matrix, and void, respectively.

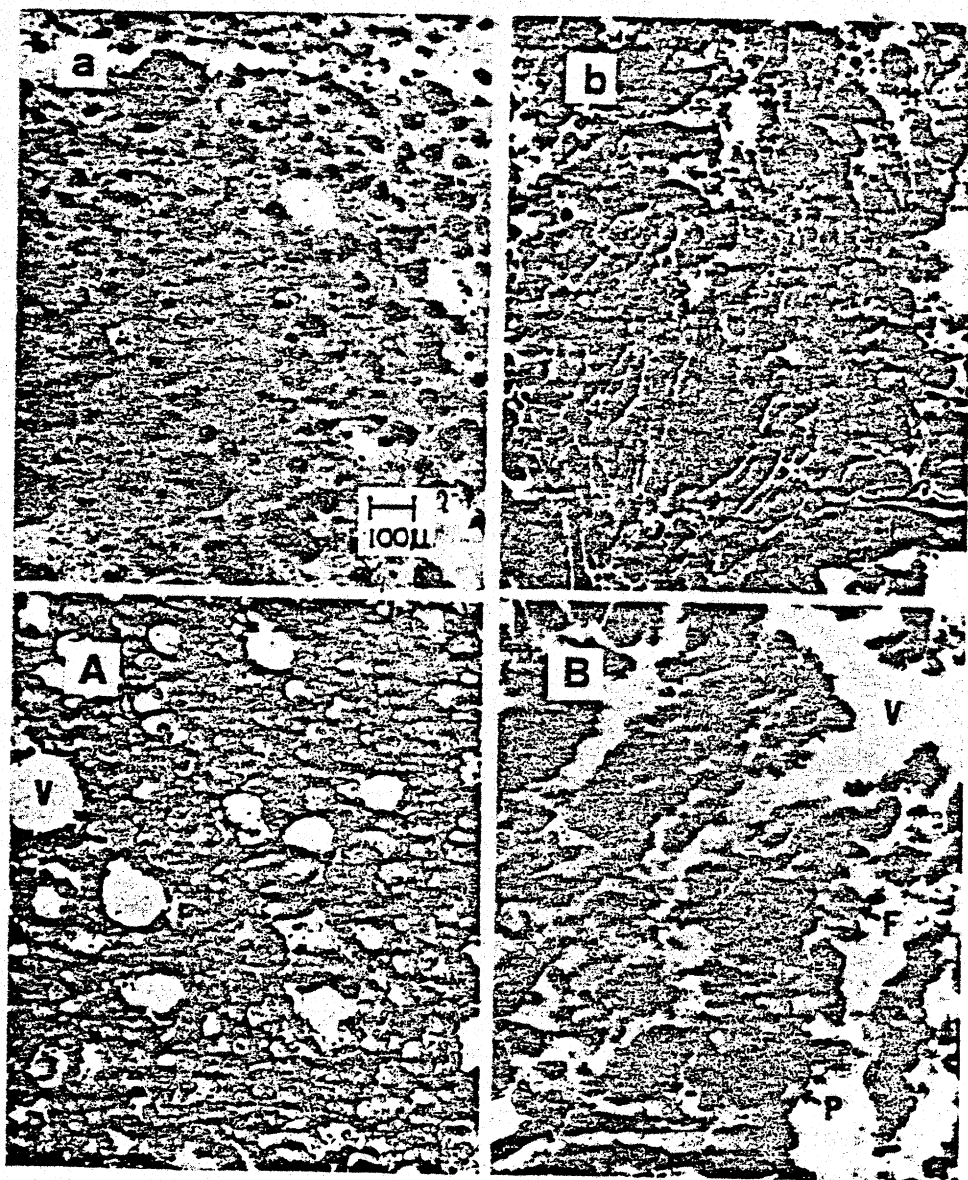
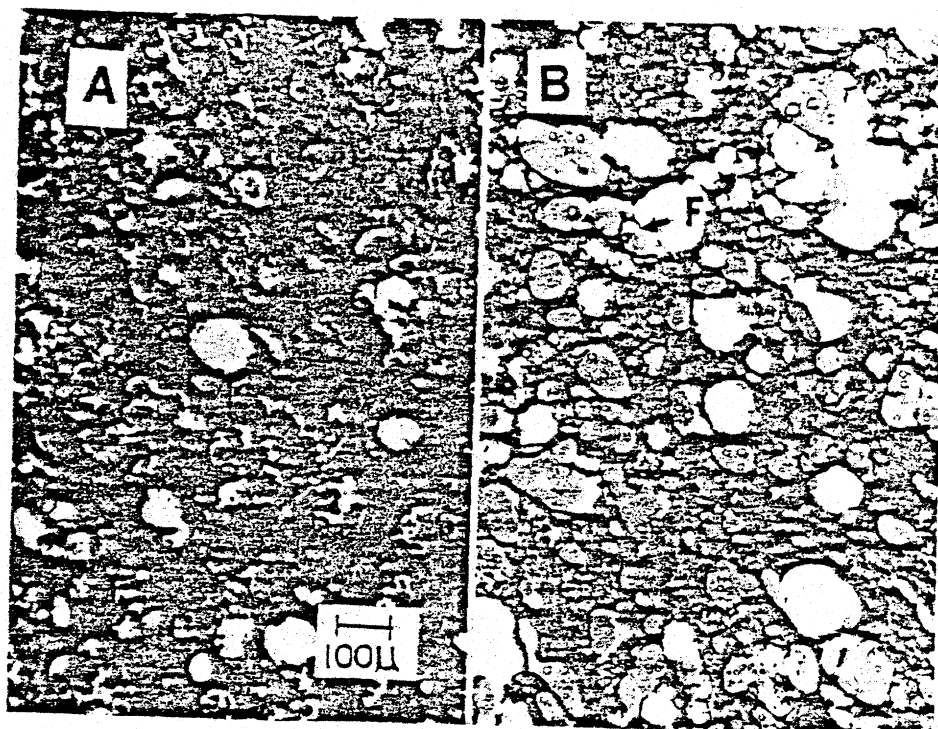


Fig. 7—Photomicrographs of uncooked emulsions prepared by adding hard fat to the batter of 26°C : (A) plastic fat of $3.0\text{ cm}^{-1}\text{ H1}$ and (B) rendered beef kidney fat of $2.1\text{ cm}^{-1}\text{ H1}$, where *F* represents fat droplets.



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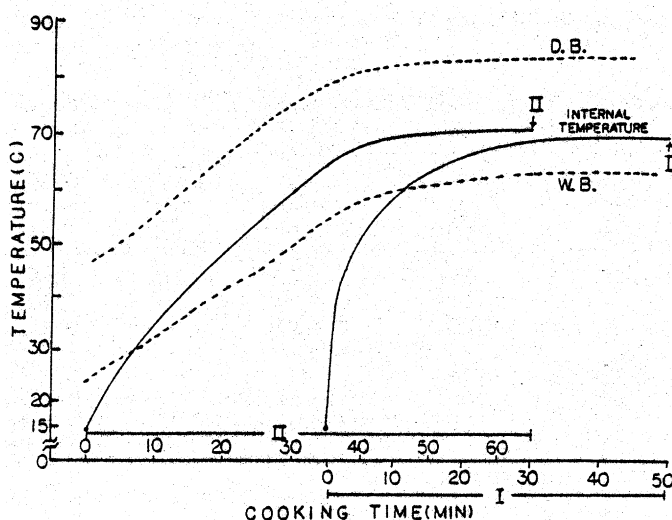


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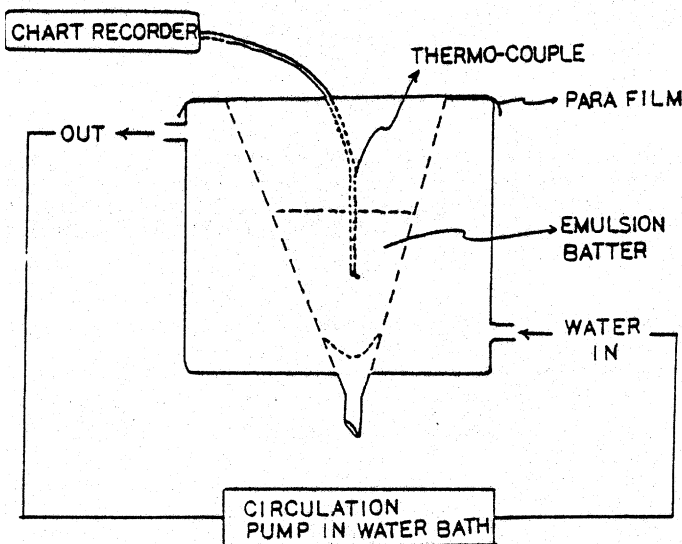


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jar and heated in a water bath at 60°C and 70°C, respectively, for 30 min. The amount of fat and water released upon heating was measured by transferring into a 10 ml graduated cylinder in order to determine a retention. The percent retention was used as a thermal stability index. Moisture contents were determined following the AOAC oven drying method (1975). Fat contents was determined following the method of Bligh and Dyer (1959) with some modifications where approximately 10g of sample was homogenized in a 100 ml capacity Waring Blendor for 1 min with 50 ml of a chloroform-methanol mixture (1:1, v/v). The 5 ml of chloroform layer was transferred into a vial and allowed to evaporate. The resulting residue was used as a crude fat residue. The internal temperature of the batter at which a release of fluid commenced was measured by monitoring changes in the internal temperature. As shown in Fig. 3 approximately 20g of batter was placed in a water-jacket funnel and a thermo-couple probe (copper-constantan) was placed in the center of the batter by resting the tip at 1/3 of the specimen height. Temperature of the water passing through the jacket was maintained at either 60° or 70°C by circulating water of equivalent temperature at a rate of 30 ml/sec. The temperature at which the batter yielded the first drop of fluid was used as a commencing temperature.

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interfacial film, the protein matrix is the one which keeps the fat from coalescing by restraining a mobilization of fat once a localization of fat is completed. Emulsion breakdown at high chopping temperature is a result of an increased mobility of fat after softening. Beyond the softening point, the mobility of fat overcomes the ability of the protein matrix to maintain uniform fat distribution by restraining fat coalescence. This apparently does not support the earlier study by Hansen (1960) who reported that the protein denaturation due to a temperature rise during chopping caused a matrix breakdown which permitted fat separation during cooking. Such destabilized emulsions were restored by addition of dry ice (Helmer and Saffle, 1963). From our results, stabilization after addition of dry ice can be explained by fat hardening which permits uniform fat distribution. It is unlikely that such stabilization occurred as a result of the physical changes of the protein gel matrix due to a temperature drop.

The mobility of fat is inversely related to the hardness of fat. It increases as the fat softens and reaches a maximum at 0% solid fat content. The mobility of fat at a particular chopping temperature affected the fat distribution pattern which in turn determined the fat stabilization in a protein matrix. The distribution pattern and the shape and size of the localized fat were found to be affected primarily both by the hardness of added fats and by the physical state of fats that changes during thermal transition occurring as a result of a temperature rise during chopping as seen in Fig. 5 and 7 series. The fat dispersion affected by melting characteristics was previously reported by Ackerman et al. (1971) who observed that pork fat was dispersed more thoroughly than beef fat.

On the basis of our results it is suggested that for the gel-type emulsions, fat stabilization can be achieved if the uniform fat dispersion is obtained by using fats of appropriate hardness under a given comminution system.

In Table 1, a similar effect on the apparent viscosity of emulsions is shown by chopping temperature and hardness of fat. The apparent viscosity of the emulsions prepared either at 16°C or with fat of 0.87 cm⁻¹ HI (equivalent to 30% solid fat) was close to that of the batter prepared without fat. This may indicate that these emulsions had stable and cohesive protein-fat matrices and a consistency similar to that of the batter prepared without fat. However, the protein-fat matrix gradually lost its continuity with an increase in either chopping temperature or the softness of fat. As a result, its matrix no longer became cohesive. This was clearly reflected by decreases in the apparent viscosity or a loose consistency. Townsend et al., (1971) reported a similar observation in which viscosities expressed in terms of Brookfield reading tended to decrease with increasing the chopping temperature.

A discontinuity of protein matrix as seen in Figure 5-B and 5-C was responsible for marked reduction in the retention of both water and fat (Table 2). Results also showed that heating at 70°C significantly reduced retention of both water and fat, compared to heating at 60°C. A reduction in retention became quite noticeable as the protein matrix lost its continuity with an increase in the chopping temperature. In the same table, compared to a slow heating at 60°C, a fast heating at 70°C significantly decreased the commencing temperatures, making emulsions more susceptible to thermal destabilization. This may explain why more frequent emulsion breakdowns during smoking are caused by cooking at high temperature and humidity which increase heating rate.

A discontinuity of protein matrix also resulted in a drastic weakening of compression strength as shown in Table 3. Fast heating significantly ($P < 0.05$) increased compressive force of the products that were comminuted to either 16°C or 21°C. However, there were no differences

in heating effect on the compressive force of the batter that was chopped to 26°C. This is obviously due to a complete breakdown of protein matrix as a result of the fat channel formation. A drastic reduction in compressive force was observed with the batter chopped to 26°C. This clearly reflects a complete disruption of matrix. No significant difference in the failure point was observed between products cooked at slow and fast heating rates. As the chopping temperature rose to 21°C, the failure points moved toward the upper half portion (0.49) of the specimens. In general, as a chopping temperature increased, the cooked products gradually lost elastic deformation and became less recoverable, as seen in Table 3. This result may indicate that the protein matrix became less cohesive and gradually lost elasticity as a result of increasing discontinuity.

In a comminuted fish muscle system, no coalescence of fat occurred to the emulsions prepared with a soft fat (equivalent to 0.25 cm⁻¹ HI). Instead, fat was finely dispersed throughout matrix (Lee and Abdollahi, 1981). In comparison with a beef muscle system, this appears to be quite contradictory. In earlier work by Townsend et al. (1971), a stable meat emulsion was prepared with cotton

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Table 1—Effect of chopping temperature and fat hardness on the apparent viscosity of emulsions^a

	Chopping temp (°C)	Apparent viscosity ^b (Poise)	Hardness (HI cm ⁻¹)	Solid fat content (%)	Apparent viscosity (Poise)
W/Fat	16	459a	0.87	30	436d
	21	357b	0.54	20	382e
	26	220c	0.25	10	218f
W/O Fat	16	430			

^a Values with different letters are significantly different within each series of treatments for chopping temperature and fat hardness, respectively ($P < 0.001$), except for the batters with and without fat at 16°C ($P < 0.02$).

^b The apparent viscosity was measured at 20 rpm which gave a computed shear rate value of 2.08 sec⁻¹.

Table 2—Effect of chopping temperature on thermal stability of meat emulsions^a

Emulsion temp (°C)	Retention (%)				Commencing temp (°C)	
	Water		Fat Heating Temp (°C)		60	70
	60	70	60	70		
16	96.2a	89.7d	96.2a	95.4a	56.5a	51.5a
21	84.1b	74.3e	75.3b	62.7c	44.0b	42.5b
26	77.2c	71.0e	59.2c	49.1d	38.0c	32.5d

^a All measurements were made in triplicate. Values with different letters are significantly different within each series of measurements at $P < 0.05$.

Table 3—Effect of chopping temperature on the mechanical properties of cooked meat emulsions^a

Emulsion temp (°C)	Compressive force (kg)		Failure point (X/L) ^b		Plastic (%) Deformation	
	I	II	I	II	I	II
16	4.9a	4.2a	0.55a	0.50a	4.16a	5.0d
21	3.6b	2.9d	0.49b	0.49	8.33b	9.16e
26	1.5c	1.3c	0.43c	0.46	12.5c	10.8e

^a I and II represent fast and slow heating, respectively (see Fig. 2). Values with different letters are significantly different within each series of measurements at $P < 0.05$.

^b X/L, where X is the distance to the point at which failure occurred, and L is specimen height.

oil. However, this study did not give information on how cotton oil was stabilized in comminuted meat matrix. The answer to this question would undoubtedly help clarify the current meat emulsion theories.

The fish muscle contains less nonmyofibrillar protein fractions such as collagen and connective tissue which make the matrix less cohesive and less uniform. Due to being less cohesive than the comminuted fish muscle matrix, the comminuted beef muscle matrix allowed the soft fat to run loose through the interstitial spaces, irrespective of a formation of the interfacial film. This observation suggests the importance of the cohesiveness of the matrix as well as the hardness of fat in a fat stabilization in the gel-type emulsion.

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